McCormick et al Serial Number 09/539,382 Page 2 of 11

## AMENDMENTS TO THE CLAIMS;

This listing of claims will replace all prior versions and listing of the claims in the application:

## **LISTING OF THE CLAIMS:**

Claims 1-53. (canceled)

Claim 54. (presently amended) A polynucleotide <u>comprising a nucleic acid</u> sequence encoding a polypeptide epitope of a B-cell lymphoma surface immunoglobulin antigen useful as a tumor-specific vaccine in a subject with a tumor or at risk of developing a tumor, encoded at least in part by a nucleic acid in the cells of said tumor, and a nucleic acid sequence promoting expression of said polypeptide in a plant cell or plant and a nucleic acid sequence inducing transient replication of said polynucleotide in the cytoplasm, which polypeptide:

- (a) includes an epitope or epitopes unique to, or overexpressed by, cells of said tumor, thereby distinguishing said tumor from all other tumors (i) of the same or different histological type, (ii) in said subject or in another member of said subject's species;
- (b) is produced in a plant cell or plant that has been transformed or transfected with said nucleic acid derived from said tumor of said subject;
- (c) is obtainable from said plant cell or plant in correctly folded form, without a need for denaturation and renaturation and mimics said epitope or epitopes in their native form; and
- (d) is capable of inducing an immune response in a mammal, including said subject, so that administration of said polypeptide results in an antibody or cell-mediated immune response to said epitope or epitopes.

Claim 55. (canceled)

McCormick et al Serial Number 09/539,382 Page 3 of 11

Claim 56. (previously presented) The polynucleotide of claim 54 wherein said polypeptide is produced transiently in said transformed or transfected plant.

Claims 57-59. (canceled)

Claim 60. (previously presented) The polynucleotide of claim 54 said polypeptide comprising two V region domains of said immunoglobulin.

Claim 61. (previously presented) The polynucleotide of claim 60 wherein said two domains of said polypeptide are at least part of the  $V_H$  and at least part of the  $V_L$  domains of said immunoglobulin.

Claim 62. (previously presented) The polynucleotide of claim 61 wherein said part of the  $V_H$  region of said polypeptide includes at least one complementarity-determining region (CDR).

Claim 63. (previously presented) The polynucleotide of claim 62 wherein said CDR of said polypeptide is CDR2.

Claim 64. (previously presented) The polynucleotide of claim 61 wherein said polypeptide is a two-domain single-chain antibody (scFV) that includes said at least part of the  $V_H$  and  $V_L$  domains

Claim 65. (canceled)

Claim 66. (previously presented) The polynucleotide of claim 64 wherein said domains of said polypeptides are linked by an amino acid linker that

- (a) has between one and about 50 residues;
- (b) consists of between one and 12 different amino acids, and

McCormick et al Serial Number 09/539,382 Page 4 of 11

- (c) facilitates secretion and correct folding of said polypeptide to mimic the tumor epitope in its native form in or on said tumor cell.
- Claim 67. (previously presented) The polynucleotide of claim 66 wherein the linker of said polypeptide is a member of a randomized library of linkers that vary in size and sequence, and said library is encoded by nucleic acid sequences consisting of a repeated pattern of degenerate repeated triplet nucleotides having the following requirements;

LSP Accounting

- (i) position 1 of each repeated triplet cannot be the same nucleotide as position 2 or the repeated triplet;
- (ii) position 2 of each repeated triplet cannot be the same nucleotide as position 3 of the repeated triplet; or
- (iii) position 1 of each repeated triplet cannot be the same nucleotide as position 3 of the repeated triplet.

Claim 68. (canceled)

Claim 69. (previously presented) The polynucleotide of claim 66, wherein in said linker of said polypeptide,

- position 1 of each repeated triplet is deoxyadenosine or deoxyguanosine; (i)
- (ii) position 2 of each repeated triplet is deoxycytidine or deoxyguanosine; and
- (iii) position 3 of each repeated triplet is deoxythymidine.

Claims 70-71. (canceled)

- Claim 72. The polynucleotide of claim 54, wherein said (previously presented) immune response is a protective anti-tumor immune response.
- Claim 73. The polynucleotide of claim 54, wherein on (previously presented) administration to a mammalian host, including said subject, said polypeptide induces a polyclonal anti-idiotypic antibody response or a cell mediated immune response.

McCormick et al Serial Number 09/539,382 Page 5 of 11

## Claims 74-75. (canceled)

Claim 76. (previously presented) The polynucleotide of claim 73, wherein said administration comprises subcutaneous immunization with at least about  $15\mu g$  of said polypeptide antigen three times each about two weeks apart.

Claim 77. (previously presented) A polynucleotide encoding a two domain single chain antibody (scFv) wherein a first domain is linked to a second domain by an amino acid linker that

- (ii) has between one and about 50 residues;
- (iii) consists of between one and 12 different amino acids,
- (iv) facilitates secretion and correct folding of said polypeptide to mimic the tumor epitope in its native form in or on said tumor cell,
- (v) is a member of a randomized library of linkers that vary in size and sequence, and said library is encoded by nucleic acid sequences consisting of a repeated pattern of degenerate repeated triplet nucleotides having the following requirements;
  - a) position 1 of each repeated triplet cannot be the same nucleotide as position
    2 of the repeated triplet;
  - position 2 of each repeated triplet cannot be the same nucleotide as position
    3 of the repeated triplet; or
  - position 1 of each repeated triplet cannot be the same nucleotide as position
    3 of the repeated triplet.

Claim 78. (previously presented) The polynucleotide of claim 77, wherein in said linker of said polypeptide,

- (i) position 1 of each repeated triplet is deoxyadenosine or deoxyguanosine;
- (ii) position 2 of each repeated triplet is deoxycytidine or deoxyguanosine; and
- (iii) position 3 of each repeated triplet is deoxythymidine.

McCormick et al Scrial Number 09/539,382 Page 6 of 11

- Claim 79. (previously presented) The polynucleotide of claim 78 wherein said scFv includes at least part of the  $V_H$  domain and at least part of the  $V_L$  domain.
- Claim 80. (previously presented) The polynucleotide of claim 79 wherein said domains are those of a surface immunoglobulin epitope of a B-cell lymphoma.
- Claim 81. (previously presented) The polynucleotide of claim 54 wherein the polypeptide is capable of inducing said immune response without a need for adjuvant or other immunostimulatory materials.
- Claim 82. (presently amended) A polynucleotide <u>comprising a nucleic acid sequence</u> encoding a polypeptide epitope of a B-cell lymphoma surface immunoglobulin antigen useful as a tumor-specific vaccine in a subject with a tumor or at risk of developing a tumor, encoded at least in part by a nucleic acid in the cells of said tumor, and a nucleic acid sequence of a vector capable of transiently replicating in the cytoplasm of and promoting expression of said polypeptide in a plant cell or plant, which polypeptide:
- (a) includes an epitope or epitopes unique to, or overexpressed by, cells of said tumor, thereby distinguishing said tumor from all other tumors (i) of the same or different histological type, (ii) in said subject or in another member of said subject's species;
- (b) is capable of being produced in a plant cell or plant that has been transformed or transfected with said nucleic acid derived from said tumor of said subject;
- (c) is obtainable from said plant cell or plant in correctly folded form, without a need for denaturation and renaturation and mimics said epitope or epitopes in their native form; and
- (d) is capable of inducing an immune response in a mammal, including said subject, so that administration of said polypeptide results in an antibody or cell-mediated immune response to said epitope or epitopes.
- Claim 83. (previously presented) The polynucleotide of claim 82 wherein said vector is a plant virus.

McCormick et al Serial Number 09/539,382 Page 7 of 11

Claim 84. (previously presented) The polynucleotide of claim 82 wherein said polypeptide is produced transiently in said transformed or transfected plant.

Claim 85. (previously presented) The polynucleotide of claim 82 wherein said vector contains a subgenomic promoter capable of promoting expression of said polypeptide.

Claim 86. (previously presented) The polynucleotide of claim 82 wherein said polypeptide is a two-domain single chain antibody (scFv) that includes said at least part of the V<sub>H</sub> and the V<sub>L</sub> domains.